

# Sampling of Vehicle Emissions for Chemical Analysis and Biological Testing

by Dennis Schuetzle\*

Representative dilution tube sampling techniques for particulate and gas phase vehicle emissions are described using Teflon filter media and XAD-2 resin. More than 90% of the total gas ( $C_8-C_{18}$ ) and particulate direct acting Ames assay mutagenicity (TA 98) was found in the particulate phase. The gas and particulate phase material was fractionated by HPLC into nonpolar, moderately polar and highly polar chemical fractions. The moderately polar chemical fraction of the particulates contained more than 50% of the direct acting Ames assay mutagenicity for the total extract. The concentration of oxygenated polynuclear aromatic hydrocarbons (oxy-PAH) and nitrated PAH (nitro-PAH) identified in the moderately polar particulate fractions are given. Nitro-PAH account for most of the direct-acting (TA 98) Ames assay mutagenicity in these moderately polar fractions. Reactions and kinetic expressions for chemical conversion of PAH are presented. Chemical conversion of PAH to nitro-PAH during dilution tube sampling of particulates on Teflon filters and gases on XAD-2 resin is a minor problem (representing 10-20%, on the average, of the 1-nitropyrene found in extracts) at short (46 min) sampling times, at low sampling temperatures ( $42^\circ C$ ), and in diluted exhaust containing 3 ppm  $NO_2$ . Particulate emissions collected from dilution tubes on filter media appear to be representative of what is emitted in the environment as based upon a comparison of highway and laboratory studies.

## Introduction

Incomplete combustion of fuel in an engine results in the formation of a complex mixture of gaseous and particulate exhaust. The concentration of chemical species in vehicle exhaust is a function of a number of complex factors including engine type, fuel and oil consumption, and engine operating conditions. Measurement of PAH from spark-ignition gasoline engines first appeared in the literature in the fifties (1). Hoffman (2) and Begeman (3) and co-workers reported a series of studies in the early sixties on the measurement of PAH and their associated biological effects in gasoline engine exhaust.

In the past few years, exhaust measurements from diesel engines have received increasing attention because of the projected increasing usage of diesel engines in passenger cars. Concerns over

possible health effects associated with diesel particulates has prompted extensive studies on biological testing and analytical measurements. A number of new findings concerning exhaust PAH emissions have also emerged from these investigations.

Up until as recently as 1978, it was believed that the nonpolar PAH were primarily responsible for the tumor producing activity observed for exhaust particulates in laboratory animal testing. In 1978, research by Huisinigh (4) and later by others using the Ames test (5-7) showed that most of the direct-acting Ames mutagenicity in diesel as well as gasoline engine exhaust particulate extract is concentrated in the moderately polar and highly polar chemical fractions. Although the relationship between Ames mutagenicity and carcinogenicity of these samples has not been established, this work has pointed to the need for identifying the chemical components which are responsible for this mutagenicity.

The chemical analysis of these moderately polar and highly polar fractions has since been under

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extensive investigation in a number of laboratories which has resulted in the identification of various oxygenated-PAH (oxy-PAH) derivatives, mostly in diesel particulate extract. Initial work on the identification of oxy-PAH in these samples was reported by Erickson (8) for the identification of alkyl-9-fluorenones and by Rappaport (9), who identified cyclopenta(c,d)pyrene dicarboxylic acid anhydride in a relatively high mutagenic fraction of a diesel particulate extract. Pitts (9,10) and Lee (11) reported the facile oxidation of PAH during ambient air and exhaust sampling and analysis, respectively, and the identification of PAH reaction products. Schuetzle et al. (12,13) found that the most mutagenic chemical fractions of diesel particulate extracts contain primarily oxygenated PAH derivatives including compounds with hydroxy, ketone, quinone, carboxaldehyde, acid anhydride, dihydroxy, and acid substituents on parent PAH. Lee (11) and Schuetzle (12) initially demonstrated the presence of nitro-PAH in diesel particulates. Subsequent work by Pederson (13), Gibson (14), Tejada (15), Schuetzle (16-19), Peterson (20) and Xu (21) have confirmed the presence of nitro-PAH in diesel particulates.

The identification of nitrated and oxygenated PAH is of crucial importance since some of these species, e.g., 1-nitropyrene, are known to be highly mutagenic (22) in the Ames assay, and the biological activities of most of the others are still unknown. The ease of oxy-PAH formation from the oxidation, sulfonation and nitration of PAH also raises the question regarding the origin of these species, i.e., whether they are produced as "native" products during the engine combustion process, in the tail-pipe, or instead, formed as the result of chemical conversion to produce artifacts during the sampling or analysis procedures. The extent to which these artifacts may affect the results of subsequent chemical or biological studies is a question that also requires the development of analytical methods for oxygenated PAH measurements.

Emphasis in this report is placed on the development of methods for the representative sampling and chemical and biological analysis of PAH and oxygenated PAH in vehicle exhaust. The major objectives of this paper are to review previous work on the development and validation of analytical techniques for chemical analysis and Ames mutagenicity tests and to present new data which can be used to determine (1) the distribution of direct acting Ames mutagenicity (TA98) between the gas and particulate phases, (2) the concentration of PAH derivatives identified in the moderately-polar particulate fractions, (3) the major reactions and related kinetic expressions responsible for

chemical conversion of PAH during sampling, (4) the major chemical species responsible for the direct acting Ames mutagenicity of the particulates and (5) if measurements made in the laboratory are representative of what is emitted from vehicles on the road.

## Experimental Approaches

### Sampling of Vehicle Exhaust

The emissions from vehicle exhaust include both gaseous and particulate phases. The exhaust temperature is very high and a cooling step is typically required before sample collection. Most of the PAH with five or more rings and hydrocarbons with more than 18 carbon atoms are expected to be particulate-associated as the exhaust is cooled to near ambient temperatures. For the more volatile organics concurrent sampling of both gases and particulates is required to insure their efficient collection.

Two types of approaches have been used for the sampling of exhaust PAH. The first involves the use of isokinetic sampling in a dilution tube while the second involves condensation of raw exhaust by passing through a cooling system. The vehicle or engine is operated on a chassis or engine dynamometer with controlled speed and load, while monitoring regulated emissions such as hydrocarbons, carbon monoxide, carbon dioxide and oxides of nitrogen (23-24).

Dilution-tube sampling of vehicle emissions is widely used in the U.S. (23-27). The technique is designed to simulate as closely as possible the dilution of vehicle exhaust as it would occur during the first minute or two in ambient air from vehicles on the road. Figure 1 describes schematically the research dilution tube system used in our laboratory (27). The system includes an exhaust inlet manifold and mixing baffle, a dilution tube and various downstream probes and equipment. The dilution tube receives the exhaust as input, and supplies a known and reproducible quantity of filtered and humidity- and temperature-controlled ambient air for rapidly diluting and cooling the exhaust. The mixed stream flows at a measured volume rate through the dilution tube where fixed proportions are withdrawn from the tube isokinetically into the sampling devices.

A direct exhaust sampling method has also been used by Grimmer (28) and others (3,29-31). The exhaust is cooled in one or a series of successively colder traps with a large cooling surface. The cooled exhaust is then allowed to pass through a filter for any untrapped particles. Both the condensed mate-

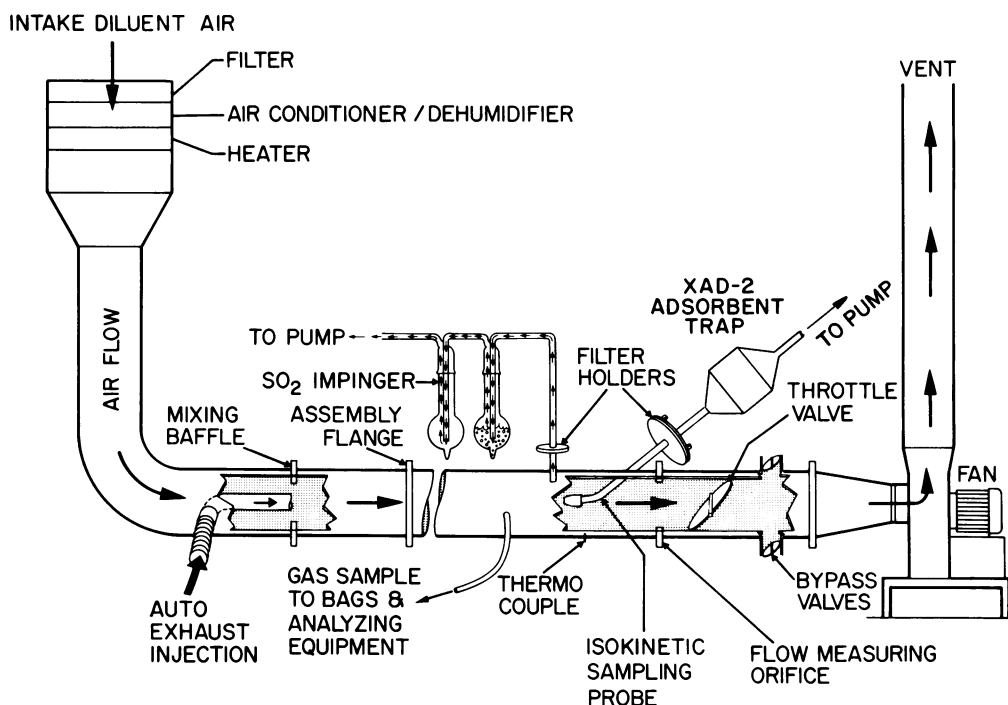


FIGURE 1. Dilution tube used for the monitoring and collection of gas and particulate-phase vehicle emissions. On-line research type analyzers include Fourier transform infrared spectroscopy, mass spectrometry and photoacoustic spectroscopy.

rial in the traps and the filter-collected particulates are analyzed without any attempt to distinguish between the two phases.

**Particulate Sampling.** Particulates emitted from vehicles may be collected using filters or electrostatic precipitators (32). Filter collection is the most widely used method. Impingers and impactors are not suitable when used alone because of their poor efficiency for collecting submicron particles generated from internal combustion engines, but may be used in series with backup filters. Various types of filter media were used in this work to determine the effect of filter composition on PAH reactions. Pure Teflon filters are preferred because of their high collection efficiency, inertness, and hydrophobicity.

**Gaseous Samplers.** Polymeric absorbents have been used widely for the sampling of gas-phase material in air and stationary-source emissions. The commonly used polymer materials include Tenax, Chromosorb 102 and XAD-2, and their applications have been described by Pellizzari (33), Jones (34) and Schuetzle (35).

A high-volume XAD-2 trap (6 in. diameter  $\times$  2 in. long, Fig. 2) was used in this work (36) for the collection of gas-phase PAH from the dilution tube. Before use, the XAD-2 polymer beads (20–50 mesh) were washed first with distilled water to remove

inorganic ions and then washed with methanol followed by dichloromethane to remove the organic contaminants. To further prevent the adsorption of impurities from air, the polymer beads were stored in methanol solvent until use. It was found that a 5–7 cm deep XAD-2 bed was sufficient for the collection of three and four ring PAHs with high efficiency even at a flow rate up to 0.8 m<sup>3</sup>/min.

## Chemical and Biological Analysis

**Extraction.** The soluble organic fraction (SOF) of the particulates which contain the organic material was separated from the exhaust particulates using organic solvent extraction procedures. Extraction was carried out by use of a Soxhlet apparatus (37). The qualitative and quantitative nature of the extracted organics was influenced by the types of solvent, the temperature and the time used for extraction.

We have previously reported (30) that Soxhlet extraction gives slightly higher mass and <sup>14</sup>C-BaP [benzo(a)pyrene] recovery than the ultrasonication extraction, that Soxhlet extraction for at least 6 hr (60 cycles/hr) is required for maximum recovery of BaP from diesel particulates collected on 47 mm Pallflex (Teflon-impregnated alkali-treated quartz fiber) filters and that binary solvent systems using

an aromatic solvent and alcohol gave the highest recovery of mass and  $^{14}\text{C}$ -BaP when compared to most other solvent systems. The gaseous PAH collected on porous polymer traps were separated by solvent extraction procedures since the PAH were not easily removed using thermal desorption. After sampling, the XAD-2 was placed in a large thimble made from stainless steel screens and extracted with pentane followed by methanol in a Soxhlet apparatus or extracted using a procedure described by Jones (34). After extraction, the volume was reduced to approximately 5 mL by Kuderna-Danish evaporation. Recoveries of standard compounds were as follows: pyrene (80%), *p*-creosol (95%), toluene (85%) and *N*-nitrosodiethylamine (35%) for samples collected at 50% RH and 30 °C.

**Fractionation.** The separation of the SOF into individual compounds is very difficult since a large number of compounds are present. Open-column liquid chromatography and liquid-liquid separation procedures (37) have been the most widely used methods for the fractionation of complex environmental particulate samples according to chemical functionalities.

The polymer adsorbent extracts were fractionated using a normal phase silica gel LC column. Ether (1%) in hexane was used to elute the nonpolar fractions. Ether/hexane (1/1) was used to elute the moderately polar fraction and MEOH was used to elute the highly polar fraction. The fractions were concentrated to approximately 1 mL by using Kuderna-Danish distillation and exchanged with DMSO before final removal of the solvent.

Normal-phase HPLC procedures were used for the fractionation of the diesel particulate extracts as described previously (17,30,38). Limited recovery studies have shown that there is little degradation or loss of certain nonpolar and moderately polar organic species on the column. No recovery studies have been undertaken on highly polar organic species. However, greater than 90% of the mass and 70–103% of Ames *Salmonella typhimurium*-active material (TA 98, -S9) injected into the column has been recovered. This variability in recovery of the Ames-active material appears to be mostly dependent on how the dose-response curves are interpreted.

**Chemical Analysis.** The complexity of vehicle emissions is compounded by the presence of unburned fuel and motor oil and by the presence of a multitude of oxidation products formed from incomplete combustion. In all, there are probably several thousand components present in vehicle exhaust which range in concentration from low ppb to high ppm levels. A typical sample size collected from a dilution tube is on the order of tens or hundreds of

milligrams. Such limited sample sizes dictate the need for sensitive analytical methods.

The major techniques employed for the analysis of organic constituents in diesel exhaust include HPLC (19,37), TLC (13), MS/MS (18), and GC/MS (8,11,12,17,18,35,39). Computer controlled GC/MS (GC/MS/COM) is the most selective and sensitive analytical technique available for these analyses (19). Both GC/MS/COM and MS/MS techniques were used for the results presented in this paper.

**Mutagenicity Testing.** The Ames *Salmonella typhimurium* test (40) was used to help establish priorities among HPLC and LC fractions for chemical analysis and to determine the contribution of an individual compound to total sample mutagenicity. The strains TA 98, TA 100 and TA 1538 were used in these studies without activation (S9) since accurate quantitative procedures with S9 have not been developed.

The percentage contribution  $P$  of an individual compound to the mutagenicity of a particulate extract was determined by using Eq. (1):

$$P(\%) = 10^{-4} (C_x)(M_x)/(M_E) \quad (1)$$

where  $C_x$  is the concentration of the component  $X$  in the extract in ppm and  $M_x$  and  $M_E$  are the slopes in revertants/ $\mu\text{g}$  of the linear portion of the dose-response functions for purified  $X$  and for the particulate extract  $E$ , respectively. Since  $M_x$  and  $M_E$  depend on the bacteria population (41), the Ames assay was carried out to ensure that these populations were the same.

It was found that certain reactive compounds, e.g., benzo(a)anthracene, isolated in HPLC fractions undergo oxidation during extended storage at 0°C. Therefore, bioassays were usually undertaken as soon as possible after fractionation. The mutagenicity of each HPLC fraction ( $A_n$ ) and the total extract ( $A_E$ ) were determined. From these results, the percentage of Ames activity recovered from the HPLC was determined from the Eq. (2)

$$A_{\text{HPLC}} = \left[ \sum_{n=1}^{n=x} A_n(W_n/W_E)(100) \right] / A_E \quad (2)$$

where  $W_n$  = mass of fraction  $n$  collected from the HPLC and  $W_E$  = mass of total extract injected into the HPLC. Using this procedure we found that nearly 100% of the indirect and direct acting mutagenicity was recovered from the normal-phase HPLC column.

Matrix studies were undertaken by comparing the dose response functions of the pure compound to that of the pure compound in the presence of

extract. Identical slopes indicated that the extract matrix has little effect on the mutagenicity of the pure compound (42).

The LC fractions were collected in preliminary experiments for the purpose of studying the distribution of mutagenic material between the gas and particulate phases. These fractions were tested using a spot test as a preliminary screen to estimate the level of Ames mutagenicity and the possible presence of toxicity. Strains TA 1535, TA 1537, TA 98 and TA 100 were used with and without activation. However TA 1535 and TA 1537 were unresponsive to the fractions. Therefore, TA 98 and TA 100 were used in a plate incorporation assay in duplicate (43). Stump (44) has recently reported a similar procedure for the Ames mutagenicity studies of unfractionated polymer adsorbent extracts.

The plate incorporation assay was not adequate to detect mutagenicity of compounds with high vapor pressure because this technique required incubation with the sample at 45°C. Quantitation of compounds with boiling points below this temperature are difficult to obtain accurately but semi-quantitative results have been obtained on volatile compounds such as vinyl chloride (bp -14°C) by using modified plate assay techniques (45, 46). As discussed previously, diethylnitrosamine (bp -177°C) was collected with 35% efficiency, whereas toluene (bp -111°C) was collected with 85% efficiency by using the XAD-2 and Kuderna Danish distillation techniques. Therefore, compounds of higher volatility could not be measured quantitatively. More work will be required to determine possible losses for a wide range of volatile materials.

## Results and Discussion

### Chemical Composition

Vehicle emissions contain a wide variety of inorganic (47-49) and organic materials (12,17,39,48). A comprehensive description of the chemical composition of vehicle emissions is beyond the scope of this paper. Instead, emphasis is placed on those substances which may be mutagenic or carcinogenic. The substances which may fall into that category include PAH, oxygenated PAH derivatives (oxy-PAH) and heterocyclic organic compounds.

**Gas Phase.** Many organic compounds emitted from engines are distributed between the particulate and gas phase (50-52). Table 1 gives the distribution of PAH in the particulate and gas phase from a stratified-charge engine. It is seen that substantial quantities of two- to four-ring PAHs are present in the gas phase. The distribution of organic compounds between the gas and particulate phases is

**Table 1. Partitioning of some PAH in the particulate and gas phase from diluted gasoline vehicle emissions.<sup>a</sup>**

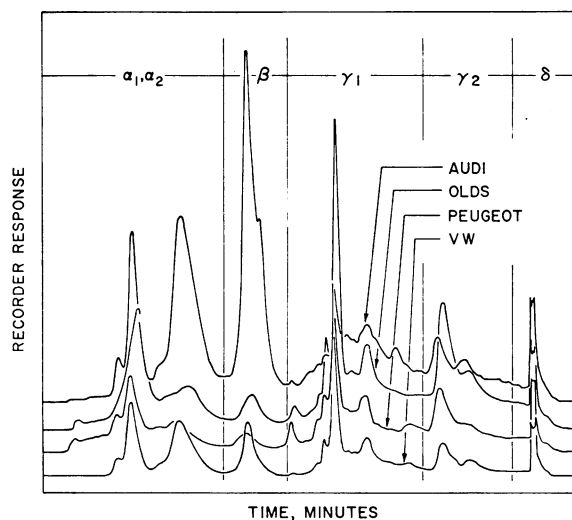
PAH	Partition coefficient <sup>b</sup>
Anthracene + phenanthrene	0.14-0.27
Fluoranthene	0.81-1.13
Pyrene	0.60-0.91
Benzo(a)anthracene + isomers	1.05-1.40
Benzo(a)pyrene	15-27
Coronene	20-32

<sup>a</sup>The collecting conditions were 37°C and 20% R.H. The exhaust was diluted 15/1 (36).

<sup>b</sup>The partition coefficient is defined as particulate/gas-phase mass ratios.

probably dependent upon a number of factors including sampling temperature and exhaust dilution ratios, but few data on the effect of these variables are available to date.

**Particulate Phase.** The HPLC/fluorescence detection procedure is not well suited for the analysis of individual components in these samples because of its inherently poor chromatographic resolution and spectral specificity. However, it can be used to compare gross chemical differences among different particulate samples. HPLC/fluorescence profiles for particulate extracts generated from several engine types are shown in Figure 2. The major peaks in each region of the chromatogram have retention times which are nearly identical with variations noted in peak intensity for each of the four diesel particulate extracts. From these data, it can be concluded that these samples may contain similar



**FIGURE 2.** Comparison of chromatograms generated from the normal-phase HPLC fractionation of diesel particulate extracts from several engine types.

chemical components but that the concentrations of these constituents are highly variable.

The regions of the chromatogram which elute nonpolar, moderately polar and highly polar compounds are arbitrarily designated ( $\alpha_1$ ,  $\alpha_2$ ,  $\beta$ ), ( $\gamma_1$ ,  $\gamma_2$ ) and ( $\delta_1$ ,  $\delta_2$ ), respectively. The distribution of fraction mass for two diesel particulate extracts is given in Table 2. The chemical constituents which have been found to be present in these fractions are summarized as follows (11,12,53).

$\alpha_1\alpha_2$  Fraction. Most of this fraction mass (90 wt-%) is due to the presence of  $C_{14}$ – $C_{35}$  aliphatic hydrocarbons and alkyl-substituted benzenes and naphthalenes. PAH and alkyl-substituted PAH ( $C_{14}$ – $C_{24}$ ) account for the remainder of the mass. The alkyl-substituted PAH are more abundant than their parent analogs.

$\beta$  Fraction. This fraction consists primarily of PAH and alkyl substituted PAH ( $C_{16}$ – $C_{26}$ ).

$\gamma_1$  Fraction. Oxygenated PAH species account for the major portion of this fraction's mass. Ketone derivatives of PAH containing three and four rings are the most abundant species. The PAH carboxaldehydes are the second most abundant species and represent substitution to PAH compounds with two to five rings. PAH quinones (three to four rings), nitrated-PAH (three to five rings) and ketone derivatives of thioxanthene and xanthone are minor constituents.

$\gamma_2$  Fraction. This fraction is comprised mostly of oxygenated PAH species. The most abundant species are anhydrides of naphthalic and methyl-naphthalic acid. Hydroxy-PAH (three to four rings), ketone- (three to four rings) and quinone- (three to four rings) PAH derivatives represent approximately half of this fraction's mass.

$\delta_1, \delta_2$  Fraction. These fractions are primarily comprised of (PAH and alkyl substituted PAH) carboxylic and dicarboxylic acids. In addition there

are N-containing heterocyclics such as acridine. PAH amines would elute in these fractions but none have been detected to date in diesel extracts.

Due to the importance of the moderately polar fractions with respect to accounting for more than half of the total particulate extract mutagenicity, extensive studies have been undertaken in this laboratory to identify and quantify components. The concentrations of PAH, nitro-PAH and oxy-PAH were analyzed using GC/MS procedures described in the Experimental Section. The results of these analyses are given in Table 3.

## Mutagenic Components

**Gas Phase.** Work was initiated in this laboratory (43) in 1977 to determine the distribution of mutagenicity in the gas (species of volatility greater than  $C_7$ – $C_8$ ) and particulate phases of vehicle emissions. Emissions from a stratified charge engine using gasoline fuel were collected, fractionated and tested with *Salmonella typhimurium* strain TA 98 by using procedures described in the Experimental Section.

Ames assay data was generated for three nonpolar (subfractions 1-3), three moderately polar (subfractions 4-6) and two highly polar (subfractions 7-8) polymer adsorbent extracts. Only subfraction 6 showed toxicity in the Ames test. XAD-2 undergoes some reaction with exhaust gases to form quinones. These substances are eluted in fraction 6 and may be responsible for the toxic reaction observed for this sample. Individual results from subfractions 1-3, 4-5 and 7-8 were composited and are presented in Table 4. The results from subfraction 6 were not included. In this manner, fractionation of the sample minimizes the toxic effects of artifacts formed from the reaction of XAD-2 with exhaust gases.

Only 3% of the total direct-acting and 5% of the total indirect-acting TA 98 mutagenicity was found in the gas-phase sample. Similar results were obtained for the TA 100 strain. Most of the gas-phase indirect-acting mutagenicity was found in the nonpolar fraction which may be due to the presence of the PAH species (see Tables 1 and 3). These results are consistent with the recent results of Stump (44), who found that the gas-phase ( $C_7$ – $C_{18}$ ) material was near background levels for gasoline-powered vehicles and was approximately ten times less active for light duty diesel emissions than the particulate extracts.

This lack of activity in the moderately polar and highly polar gas phase fractions indicates that oxidation and nitration of the three- to four-ring PAH collected on XAD-2 during sampling is negligible.

**Table 2. Weight distribution of chemical fractions generated from the HPLC fractionation of diesel particulate extracts.**

Fraction	% weight recovered	
	Sample A	Sample B
Nonpolar		
$\alpha_1 + \alpha_2$	57.4	53.0
$\beta$	1.3	1.7
Moderately polar		
$\gamma_1$	4.3	6.9
$\gamma_2$	5.0	2.3
Polar		
$\delta_1$	16.0	16.1
$\delta_2$	14.7	16.5
Total	98.7	96.5

\*HPLC chromatographic fractions as defined in Fig. 2.

**Table 3. Concentration of various PAH compounds and PAH derivatives in the nonpolar and moderately polar fractions of a diesel particulate extract.<sup>a</sup>**

Compound	Fraction concentration, ppm/1000 <sup>b</sup>
Nonpolar fractions ( $\alpha_1$ , $\alpha_2$ , $\beta$ ) <sup>c</sup>	
PAH	
Phenanthrenes and anthracenes	1.1
Methyl (phenanthrenes and anthracenes)	2.6
Dimethyl (phenanthrenes and anthracenes)	5.8
Pyrene	3.1
Fluoranthene	2.5
Methyl (pyrenes and fluoranthenes)	1.4
Chrysene	0.18
Cyclopenta(c,d)pyrene	0.03
Benzo(g,h,i)fluoranthene	0.24
Benzo(a)anthracene	0.95
Benzo(a)pyrene	0.07
Subtotal	18.0
Other PAH, heterocyclics	62.0
Hydrocarbons and alkylbenzenes	920.0
Total	1000.0
Moderately polar fractions ( $\gamma_1$ , $\gamma_2$ ) <sup>d</sup>	
PAH ketones	
Fluorenones	43.2
Methyl fluorenones	4.8
Dimethyl fluorenones	1.8
Anthrones and phenanthrones	17.9
Methyl (anthrones and phenanthrones)	17.7
Dimethyl (anthrones and phenanthrones)	14.4
Fluoranthones and pyrones	13.4
Benzanthrones	2.1
Xanthones	3.6
Methylxanthones	1.8
Thioxanthones	17.0
Methyl thioxanthones	9.3
Subtotal	147.0
PAH carboxaldehydes	
Fluorene carboxaldehydes	17.6
Methyl fluorene carboxaldehydes	3.9
(Phenanthrene and anthracene) carboxaldehydes	28.2
Methyl (anthracene and phenanthrene carboxaldehydes)	17.7
Dimethyl (anthracene and phenanthrene carboxaldehydes)	4.8
(BaA, chrysene and triphenylene carboxaldehydes)	4.4
Naphthalene dicarboxaldehydes	3.5
Dimethyl naphthalene carboxaldehydes	3.5
Trimethyl naphthalene carboxaldehydes	10.5
(Pyrene and fluoranthene) carboxaldehydes	17.3
Xanthene carboxaldehydes	6.6
Dibenzofuran carboxaldehydes	4.2
Subtotal	122.2
PAH acid anhydrides	
Naphthalene dicarboxylic acid anhydrides	31.2
Methyl naphthalene dicarboxylic acid anhydrides	11.3
Dimethyl naphthalene dicarboxylic acid anhydrides	5.0
(Anthracene and phenanthrene) dicarboxylic acid anhydrides	6.6
Subtotal	54.1
Hydroxy-PAH	
Hydroxyfluorene	14.8
Methylhydroxyfluorene	4.1
Dimethylhydroxyfluorene	16.7
Hydroxy(anthracenes and phenanthrenes)	6.3
Hydroxymethyl(anthracenes and phenanthrenes)	9.5
Hydroxydimethyl(anthracenes and phenanthrenes)	14.4
Hydroxyfluorenone	22.4
Hydroxyxanthone	14.1
Hydroxyxanthene	10.8
Subtotal	113.1

Table 3. (continued)

Compound	Fraction concentration, ppm/1000 <sup>b</sup>
PAH quinones	
Fluorene quinones	8.1
Methylfluorene quinones	6.2
Dimethylfluorene quinones	5.3
(Anthracene and phenanthrene) quinones	20.4
Methyl (anthracene and phenanthrene) quinones	22.4
(Fluoranthene and pyrene) quinones	2.1
Naptho(1,8-c,d)pyrene-1,3-dione	6.8
Subtotal	71.3
Nitro-PAH	
Nitrofluorenes	0.34
Nitro(anthracenes and phenanthrenes)	0.71
Nitrofluoranthrenes	0.05
Nitropyrenes	1.5
Methyl nitro(pyrenes and fluoranthenes)	0.25
Subtotal	2.9
Other oxygenated PAH	83.4
PAH carry-over (from $\alpha_1$ , $\alpha_2$ , $\beta$ )	66.0
Phthalates, HC contaminants	340.0
Total	1000.0

<sup>a</sup>See the literature for experimental details concerning sample collection procedure (37) for details concerning quantitative MS analysis (18, 19, 35).

<sup>b</sup>Fraction concentration determined from the summation of the three most abundances in the mass spectra.

<sup>c</sup>Divide fraction concentration by 1.8 to determine total extract concentration. Fraction cut defined from elution time zero to the elution time of methyl coronene.

<sup>d</sup>Divide fraction concentration by 10.9 to determine total extract concentration. Fraction cut defined from elution of 2-nitrofluorene to that of 7-benz(a)anthrene.

Table 4. Relative values of Ames mutagenicity (TA 98) for particulate and gasphase components collected for gasoline stratified-charge engine emissions.

Chemical fractions	Relative mutagenicity (Mutagenicity, rev/mg) <sup>a,c</sup>			
	Particulate extract <sup>b</sup>		Adsorbent extract <sup>b</sup>	
	-S9	+S9	-S9	+S9
Nonpolar ( $\alpha_1$ , $\alpha_2$ , $\beta$ )	<0.3 ( <81)	4.2 (1134)	0.4 (108)	1.0 <sup>a</sup> (270)
Moderately polar ( $\gamma_1$ , $\gamma_2$ )	15.2 (4050)	14.0 (3780)	<0.3 ( <81)	<0.3 ( <81)
Polar ( $\delta_1$ , $\delta_2$ )	8.1 (2187)	1.8 ( 486)	0.3 ( 486)	<0.3 ( <81)
Total activity	23.6 (6370)	20.0 (5400)	0.7 (189)	1.0 (270)

<sup>a</sup>The mutagenicity values in total revertants for each fraction are normalized to the total revertants observed for the adsorbent extract.

<sup>b</sup>See Table 1 for PAH compositional data.

<sup>c</sup>All mutagenicity values are blank corrected; 2-nitrofluorene = 167 rev/mg.

These data are consistent with the chemical reactivity model experiments described in the next section.

Egeback (56) collected gas-phase emissions after filtering of the particulates by using a condensation technique. The direct-acting Ames mutagenicity of the condensate was found to be much greater than that of the particulates. The reasons for the difference between Egeback's data and our results are not known. Further studies will be required to determine if artifact formation of mutagenic species may occur in the condensation procedure or if chem-

ical reactions or physical losses in the XAD-2 resin procedure result in low mutagenicity values.

There is little information available on the highly volatile components ( $C_2$ - $C_7$ ) due to the difficulty of gas-phase bioassay testing. However, a substantial body of data exists on the  $C_2$ - $C_7$  components present in diesel and spark-ignition gasoline engines (53-55). The only components identified to date in the  $C_2$ - $C_7$  range which appear to have mutagenic potential are the aldehydes and the halogenated hydrocarbons such as dibromoethane.



**Particulate Phase.** Most of the direct-acting Ames mutagenicity of the particulate extracts was found in the moderately polar fraction. These fractions ( $\gamma_1$  and  $\gamma_2$ ) represent 50–75% of the direct-acting and 20–70% of the indirect-acting mutagenicity (TA 98) for total unfractionated diesel exhaust (12,17,57). For this reason, emphasis has been placed on the identification of mutagens in the moderately polar fraction.

It is now known that a large number of nitrated-PAH (nitro-PAH) compounds are present in these samples (12,18,21). One of these nitro-PAH (1-nitropyrene) accounts for up to 30% of the TA 98 direct-acting mutagenicity (12,13,17,19,58).

Table 5 gives the concentration range or upper concentration limits for nitro-PAH in several diesel particulate extracts as determined using the MS/MS and GC/MS procedures described in the Experimental Section (19). The direct acting mutagenicity of the predominant isomer is given for TA 98. The concentration and percentage contribution of each compound to total extract mutagenicity was then calculated. It can be seen that these nitro-PAH together account for 40% or more of the total extract direct-acting mutagenicity. The most significant

mutagens identified in these samples to date are the nitropyrenes, dinitropyrenes and nitrohydroxypyrenes. The concentration of the dinitropyrene is probably low since recoveries have not been determined pending synthesis of deuterated standards (35).

## Chemical Conversion during Sampling and Analysis

The oxy-PAH found in ambient particulates may be present as a result of primary emissions, atmospheric reactions or chemical conversion (artifacts) during sampling.

Pitts (10,59) and Lane (60) first showed that PAH deposited on filters react with  $\text{NO}_2$  (probably containing traces of  $\text{HNO}_3$ ) and  $\text{O}_2$ . In these studies, Pitts (59) identified mononitro-PAH and PAH-epoxides, and Hughes (61) identified mononitro and dinitro-PAH reaction products. These results were consistent with those of Vollman (62) and Dewar (63) who showed that PAH react with nitric acid to form mononitro, dinitro, trinitro and tetranitro species. Pitts (10) found that some of these nitro-PAH

Table 5. Concentration and percentage contribution to total extract Ames direct-acting mutagenicity (TA 98) for some nitro-PAH identified in diesel particulate extracts.

Compounds <sup>a</sup>	Mutagenicity, rev/ $\mu\text{g}^b$	Concentration, ppm			Contribution to total extract mutagenicity, % <sup>c</sup>		
		A	B	C	A	B	C
Nitrofluorenes (2)	150	186.0 <sup>d</sup>	78.0 <sup>d</sup>	71.0 <sup>d</sup>	0.2	1.0	1.6
Dinitrofluorene (2,7)	2,100	4.2	6.0	$\leq 3.0$ (nd)	0.05	1.1	$\leq 0.9$ (nd)
Dinitro-9-fluorenone (2,7)	6,700	8.6	3.0	$\leq 3.0$ (nd)	0.3	3.4	$\leq 3.0$ (nd)
Nitroanthracene (9)	0.5	552.0 <sup>d</sup>	150.0 <sup>d</sup>	81.0 <sup>d</sup>	$< 0.01$	$< 0.01$	$< 0.01$
Nitropyrene (1)	2,000	2030.0 <sup>e</sup>	185.0 <sup>e</sup>	55.0 <sup>e</sup>	24.0	30.8	16.2
Nitrofluoranthenes (3,8)	28,000 (2.7/1.0)	6.3	$\leq 5.6$ (nd)	$\leq 1.7$ (nd)	1.0	$\leq 13.1$ (nd)	$\leq 7.0$ (nd)
Dinitropyrenes (1,3; 1,6; 1,8)	670,000 (1.0/1.3/2.2)	5.2 <sup>d,f</sup>	$\leq 0.5$ (nd)	$\leq 0.5$ (nd)	20.6	$\leq 27.9$ (nd)	$\leq 49.3$ (nd)
Nitromethylpyrenes (1,3; 6,1; 8,1)	270 (1.0/1.6/2.0)	146.0	46.0	16.0	0.2	1.0	0.6
Nitrohydroxypyrenes (3,1; 6,1; 8,1)	7,500 (1.0/1.5/2.0)	74.0 <sup>h</sup>	46.0 <sup>h</sup>	10.0 <sup>h</sup>	3.3	28.8	14.7
Nitrobenzo(a)pyrenes (6;3)	180 (2.8/1.0)	82.0	22.0	10.0	0.1	0.3	0.3

<sup>a</sup>Number in parentheses represents isomers found as identified by comparison of retention times with authentic standards.

<sup>b</sup>Mutagenicity value is given for a mixture with the same isomer distribution (given in parentheses) as found present in the samples.

<sup>c</sup>Extract mutagenicity of A, B, and C is 16.9, 1.2, and 0.68 rev/ $\mu\text{g}$ , respectively; % contribution calculated from Eq. (1); A, B and C are  $\text{MeCl}_2$  extracts of three light-duty diesel particulate extracts.

<sup>d</sup>Concentrations are accurate to within  $\pm 30$ –40%.

<sup>e</sup>Concentrations are accurate to within  $\pm 10$ –15%; all other concentrations are accurate to within  $\pm 50$ –100%; nd = not detected.

<sup>f</sup>Concentration determined by GC/N-selective detection.

<sup>g</sup>Total mutagenicity of mixture was due primarily to the mutagenicity of the 3,1 isomer which was  $30,000 \pm 4,000$  rev/ $\mu\text{g}$ .

<sup>h</sup>Concentration determined using MS/MS analysis (18); Distribution of 3,1; 6,1 and 8,1 isomers based upon distribution of isomers obtained from laboratory nitration of 1-hydroxypyrene.

(6-nitro-BaP, 3-nitroperylene, and 1-nitropyrene) exhibited direct-acting Ames mutagenicity.

Lee (11,64) showed that the oxidation of PAH is greatly enhanced by the direct deposition of the PAH on the quartz or glass fiber filters used in the experiments by Pitts and Lane, and that PAH deposited on Teflon filters or adsorbed in particulates are much more stable toward chemical conversion. These effects are demonstrated by the recovery of BaP during various conditions of ambient air sampling as shown in Figure 3.

In order to determine the reactivity of PAH in the presence of diesel exhaust, extensive recovery experiments have been undertaken on PAH compounds in this laboratory (11,64,65) and at Battelle-Columbus Research Laboratories (66). Figure 4 summarizes results from several of these studies using fluoranthene (F), chrysene (C), pyrene (P) and benzo(a)pyrene (B). It is seen that the PAH deposited on Teflon-coated glass filters (e-h) readily undergo reactions with diesel exhaust. However, the reaction of these PAH with diesel exhaust is greatly reduced once the PAH are adsorbed on XAD-2 (a-d) resins or on particulate matter (i-k).

The data in Figure 4 show that the PAH injected into hot diesel exhaust (300°C) are rapidly degraded (l-n). The PAH would be present in the gas phase at these exhaust temperatures (300°C). Therefore, loss of these PAH is probably the result of a gas-phase oxidation, possibly enhanced by the presence of sulfuric and nitric acids. Product analysis will be needed to help determine the possible reaction mechanisms. In this experiment the PAH are exposed to the 300°C exhaust gases for an estimated 0.2–0.3 sec before being cooled to 45°C. From these data, the half-lives of chrysene, pyrene and BaP are 0.3–0.4 sec, 0.04–0.06 sec and  $\leq 0.02$ –0.03 sec, respectively.

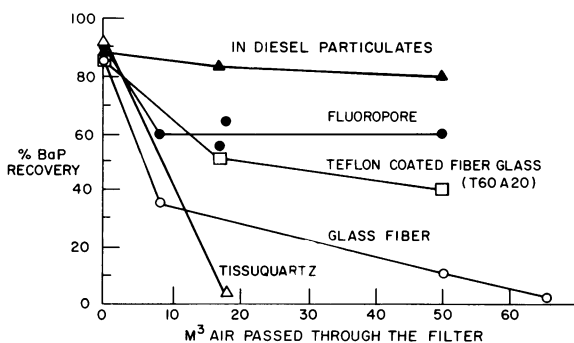


FIGURE 3. Recovery of BaP during various conditions of ambient air sampling for 27 ppm of BaP in diesel particulates on a T60A20 filter and for 500  $\mu$ g of BaP spiked on various filter media (36).

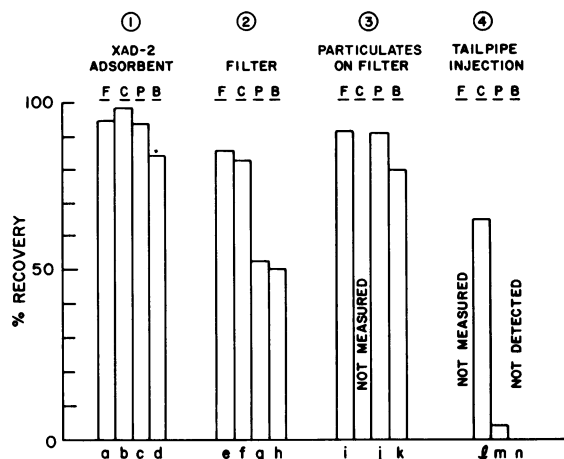
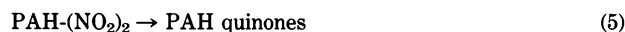
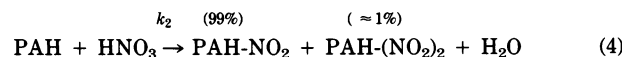
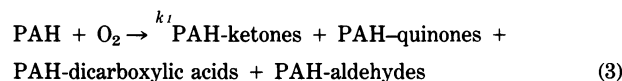
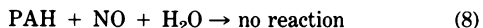
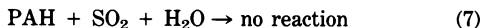
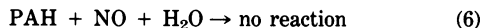


FIGURE 4. Recovery of fluoranthene (F), chrysene (C), pyrene (P) and benzo(a)pyrene (B) exposed to diluted (1/12) and filtered diesel exhaust (40°C) for 23 min from a diesel engine operating at 1500 rpm at half-load: (1) XAD-2 adsorbent was spiked with each PAH at a level of 600  $\mu$ g/g (exposed to filtered exhaust at a rate of 560 L/min); the contribution of gas-phase engine PAH to XAD-2 recoveries is 3% for P and F and 1% for C and B; (2) Teflon impregnated glass fiber filter (20  $\times$  25 cm) was spiked with 100–500 ng of each PAH; (4) C and P (100 ng each) and B (10 ng) were spiked into exhaust at 300°C. Data are from various sources (66).

It can be concluded from these results that the reaction of PAH with exhaust is very fast. Pyrene and BaP undergo several reaction half-lives before they are emitted from the tail-pipe.

We have undertaken preliminary studies in this laboratory and analyzed limited data available in the literature to elucidate the mechanisms and kinetics for the reaction of PAH in the gas and particulate phases. Figure 5 shows the HPLC chromatograms obtained from the reaction of SO<sub>2</sub> and SO<sub>3</sub> with BaP. Peak (a) is identified as 3-BaP-SO<sub>3</sub>H as based upon direct probe MS analysis and comparison of the mass spectra with that of a synthesized standard. We have previously shown that PAH readily react with NO<sub>2</sub> but not NO (11,64). We have found also that PAH react with molecular oxygen to form quinones, ketones, aldehydes and dicarboxylic acids. The products formed from these various reactions are summarized in Eqs. (3)–(8).





From these reactions, the rate expression for loss of PAH can be written as given by Eq. (9):

$$-d[\text{PAH}]/dt = k_1 [\text{PAH}][\text{O}_2] + k_2 [\text{PAH}][\text{NO}_2] + k_3 [\text{PAH}][\text{H}_2\text{SO}_4] \quad (9)$$

where the concentration of  $\text{NO}_2$  is proportional to the  $\text{HNO}_3$  concentration. We have attempted to determine the concentration of PAH sulfonic acids in diesel particulate extracts, but these species are highly labile and accurate quantitative techniques have not been developed. However, limited analysis has shown that nitrated and oxygenated PAH are more abundant than the sulfonated PAH. Using the assumption that  $k_3$  is much less than  $k_1$  or  $k_2$  and since the oxygen concentration is several orders of magnitude higher than the PAH concentration, we can reduce Eq. (9) to Eq. (10).

$$-d[\text{PAH}]/dt = k_{1p} [\text{PAH}] + k_2 [\text{PAH}] [\text{NO}_2] \quad (10)$$

where

$$k_{1p} = k_1 [\text{O}_2] \quad (11)$$

and the constant for loss of pyrene is given by Eq. (12):

$$k_4 = k_{1p} + k_2 [\text{NO}_2] \quad (12)$$

Diesel particulate emissions are known to contain up to 3% sulfates, most of which is present as sulfuric acid (67). Some of this sulfate may be formed from the adsorption of  $\text{SO}_2$  in the particulate and conversion of the  $\text{SO}_2$  to sulfate during storage (68). However, the concentration of nitric acid is unknown. Based upon previous measurements (69) and thermodynamic considerations, the concentration of nitric acid in exhaust is estimated to be 0.01–0.001 of the  $\text{NO}_2$  concentration.

The relative reactivities of fluoranthene, chrysene, pyrene and benzo(a)pyrene to nitration and photochemically induced oxidation vary considerably as shown in Table 6. These rate data are consistent with the general trend of recovery data observed in Figure 4. Some kinetic studies on the reaction of PAH with diesel exhaust and  $\text{NO}_2$  have been undertaken in this laboratory and the kinetic information obtained from these studies are used to calculate

reaction rate constants using our data and data obtained from other investigators. These data are summarized in Table 7.

The effect of re-exposing diesel particulates to filtered diesel emissions has been determined by Gibson (14). They found that the direct-acting Ames activity and concentrations of 1-nitropyrene and 6-nitrobenzo(a)pyrene in diesel particulate samples increased with re-exposure to diesel exhaust. Using their data, the rate constants for the loss of pyrene and the formation of nitropyrene are found to be  $1.9 \times 10^{-1}/\text{hr}$  and  $2.1 \times 10^{-2}/\text{hr-ppm}$  using Eqs. (12) and (13), respectively.

$$+d[\text{nitro-PAH}]/dt = k_2 [\text{PAH}] [\text{NO}_2] \quad (13)$$

In this experiment 28% and 27% of the pyrene was converted to nitropyrene and nitro-BaP, respectively.

Gorse (71) found that diesel particulates collected for long periods of time (230 min) had direct-acting mutagenicity levels approximately 50% higher than particulates collected for 23 min periods. Samples generated in those experiments from collection on T60A20 filters were analyzed for nitropyrene and pyrene concentrations using GC/MS (72) and the data are summarized in Table 7. The particulate samples generated in this experiment were collected for 23 ( $t_1'$ ) min and 230 ( $t_2'$ ) min from an engine operating at steady state. Since the exhaust gases were not prefiltered as in Gibson's experiment, the exposure time requires correction because the pyrene deposited at  $t = 230$  min has no time for further reaction and the pyrene at  $t = 0$  has 230 min to react. Using Eqs. (14)

$$\begin{aligned} t_1 &= t_1(1 - 1/e) \\ t_2 &= t_2(1 - 1/e) \end{aligned} \quad (14)$$

$t_1$  (initial) and  $t_2$  (final) are 14 and 145 min, respectively. From these data, the rate constants for loss of pyrene and formation of nitropyrene are  $9.9 \times 10^{-2}/\text{hr}$  and  $1.1 \times 10^{-2}/\text{hr-ppm}$ , using Eqs. (12) and (13), respectively. In this experiment, 34% of the pyrene was converted to nitropyrene.

The rate constants for loss of pyrene and formation of nitropyrene are approximately two times greater in Gibson's work than they are in Schuetzle's work. These variations may be due to the differences in sample collection temperatures which indicates that the rate constant for loss of pyrene approximately doubles for each  $10^\circ\text{C}$  rise in temperature. Further work on the relationship of collection temperatures to reaction rates are needed.

Jager (73) has studied the exposure of pyrene in fly ash particulates. From that data, the rate con-

Table 6. Relative reactivity of PAH for nitration and oxidation.

PAH	Ring position	Nitration(R <sub>3</sub> ) <sup>a</sup>	Oxidation(R <sub>4</sub> ) <sup>b</sup>
Fluoranthene	3	—	1.0
Chrysene	2	1.0	—4.8
Pyrene(PYR)	1	5.0	30
Benzo(a)pyrene(BaP)	6	31	40

<sup>a</sup>Reaction with HNO<sub>3</sub> (63).<sup>b</sup>Photosensitized reaction with oxygen (70).Table 7. Reactivity of pyrene and BaP in particulates exposed to NO<sub>2</sub>, SO<sub>2</sub> and diesel exhaust.

Compound	Reference	T, °C	Exposure <sup>a</sup>		Particulate source	PAH concentration, ppm <sup>b</sup>		Time, hr	Formation of			Loss of pyrene, k <sub>4</sub> (hr <sup>-1</sup> ) <sup>e,f</sup>
			NO <sub>2</sub> , ppm	SO <sub>2</sub> , ppm		Initial	Final		Δ Nitro-PAH, ppm	Nitro-PAH, k <sub>2</sub> , (hr <sup>-1</sup> ) <sup>c,d</sup>		
Pyrene	Butler (74)	25	10	—	Ethene flame	7743	6503	120	—	—		1.5 × 10 <sup>-21</sup>
Pyrene	Schuetzle (72) <sup>g</sup>	34	3	20	Diesel	251	202	2.2	16	1.1 × 10 <sup>-2</sup>		9.9 × 10 <sup>-2</sup>
Pyrene	Gibson (14) <sup>h</sup>	43	3	20	Diesel	102	72	1.5	8.2	2.1 × 10 <sup>-2</sup>		2.3 × 10 <sup>-1</sup>
Pyrene	Jager (73)	25	1.3	—	Fly ash	100	—	4.5	50	1.1 × 10 <sup>-1</sup>		—
Pyrene	Petersen (66)	300	—	—	Diesel	—	—	—	—	—		6.1 × 10 <sup>7</sup>
Pyrene	Calculated <sup>c</sup>	300	—	—	Diesel	—	—	—	—	—		1.2 × 10 <sup>7</sup>
BaP	Butler (74)	25	10	—	Ethene flame	1557	683	120	—	—		6.4 × 10 <sup>-2</sup>
BaP	Gibson (14) <sup>h</sup>	43	3	20	Diesel	7.2	3.8	1.5	1.4	5.7 × 10 <sup>-2</sup>		4.1 × 10 <sup>-1</sup>
BaP	Calculated <sup>e</sup>	300 <sup>b</sup>	—	—	Diesel	—	—	—	—	—		3.2 × 10 <sup>7</sup>
BaP	Petersen (66)	300	—	—	Diesel	—	—	—	—	—		≤8.6 × 10 <sup>7</sup>

<sup>a</sup>Concentration of constituents are given in ppm (v/v).<sup>b</sup>Concentration in particulate.<sup>c</sup>Extrapolation to 300°C from rate constant data at 34 and 43°C listed above.<sup>d</sup>Calculated from the expression:  $+d\text{Nitro-PAH}/dt = k_2(\text{PAH})(\text{NO}_2)$  with PAH and nitro-PAH concentrations in ppm by mass in the particulate and NO<sub>2</sub> concentrations in ppm by volume in the gas stream: assumes  $\Delta\text{PAH}/\Delta t = d\text{PAH}/dt$ .<sup>e</sup>Extrapolation to 300°C from rate constant data at 43°C assuming rate constant doubles for every 10°C rise in temperature.<sup>f</sup>See Eq. (12).<sup>g</sup>Exposure to unfiltered diesel exhaust.<sup>h</sup>Exposure to filtered diesel exhaust.<sup>i</sup>NO<sub>2</sub> concentration rate limiting which significantly decreases k<sub>4</sub>.

stant for formation of nitropyrene is  $1.1 \times 10^{-1}/\text{hr-ppm}$ . This constant is approximately an order of magnitude greater than that determined from the work of Schuetzle and Gibson. The reason for these differences are not known. The rate constants, k<sub>4</sub>, for the reaction of pyrene and BaP at 300 °C are  $1.2 \times 10^7$  and  $3.2 \times 10^7/\text{hr}$ , respectively, as determined from an extrapolation of the kinetic data of Gibson and Schuetzle (Table 7), and the assumption that the rate constant (k<sub>2</sub>) doubles for each 10 °C rise in temperature for BaP. These results are in reasonable agreement with the rate constant of pyrene (k<sub>4</sub> =  $6.1 \times 10^7/\text{hr}$ ) as determined from the exhaust spiking experiments (66) described previously and summarized in Table 7.

Butler (74) found that PAH imbedded in particulates react very slowly with NO<sub>x</sub>. Butler's data for the loss of pyrene and BaP exposed to 10 ppm NO<sub>2</sub> are presented in Table 7. Unfortunately, analysis was not undertaken on the reaction products. The rate constants for the loss of pyrene and BaP are

$1.5 \times 10^{-2}$  and  $6.4 \times 10^{-2}/\text{hr}$  as calculated from Eq. (10), assuming k<sub>2</sub> is small compared to k<sub>1p</sub>, since the NO<sub>2</sub> concentration as compared to the PAH concentrations is low and the reaction is therefore kinetically rate limiting.

Butler's and Gibson's data indicate that BaP is 4.0 and 1.8 times more reactive than pyrene, respectively. This data is roughly consistent with the general trend of reactivities presented in Table 6 for these species. Using the relative reaction rates given in Table 6, and assuming that only nitration and oxidation cause degradation of pyrene and BaP in diesel exhaust, the relative reactivity of BaP to pyrene is 1.9 as calculated using Eq. (15)

$$\frac{k_1[\text{BaP}]}{k_1[\text{PYR}]} = \frac{f[\text{BaP}]R_3[\text{BaP}] + (1-f)[\text{BaP}]R_4[\text{BaP}]}{f[\text{PYR}]R_3[\text{PYR}] + (1-f)[\text{PYR}]R_4[\text{PYR}]} \quad (15)$$

where  $R[\text{BaP}]$  and  $R[\text{PYR}]$  is the fraction of BaP and pyrene converted to nitro-BaP and nitropyrene, respectively;  $(1 - f) [\text{BaP}]$  and  $(1 - f) [\text{PYR}]$  is the fraction of BaP and pyrene, respectively, converted to products resulting from oxidation; and  $R_3[\text{BaP}]$ ,  $R_3[\text{PYR}]$ ,  $R_4[\text{BaP}]$  and  $R_4[\text{PYR}]$  are given in Table 6.

It can be concluded from the preceding discussion that the problem of PAH loss and nitro-PAH formation during filter sampling of diesel particulates collected on Teflon filters from diluted exhaust containing less than 3 ppm  $\text{NO}_2$  can be minimized by short sampling times of less than 23 min (1 FTP cycle) and at sampling temperatures less than  $43^\circ\text{C}$ . Under these conditions, pyrene and BaP losses would be a maximum of 10 and 19%, respectively. Chemical conversion during dilution tube sampling accounts for less than 10–20% of the 1-nitropyrene found in diesel particulate extracts.

## Comparison of Laboratory and Highway Studies

**Particulate Phase.** As described earlier, most of the data collected on vehicle emissions is from laboratory studies using dynamometer/dilution-tube measurements. The relevance of vehicle dynamometer/dilution-tube emission measurements to the measurement of vehicle emissions emitted into the atmosphere comes under question since, for example, emissions from vehicles on the road have higher dilution ratios, have longer residence times, are collected at lower temperatures, and may interact with ambient air pollutants. Therefore, it might be expected, per the discussion in the last section, that the possibility of chemical interactions during sampling would be reduced.

Pierson et al. (75) conducted experiments in 1979 in the Allegheny Mountain Tunnel located in Pennsylvania to compare particulate emissions generated from vehicles on the road with emissions generated from vehicles in laboratory dilution tube studies. Some results of that study are summarized in Table 8.

A comparison of particulate emissions from spark-ignition vehicles and heavy duty diesel vehicles was made in the Allegheny Mountain Tunnel studies. It was found that the particulate emissions from heavy duty diesels are some two orders of magnitude greater than the particulate emissions generated from spark-ignition vehicles (primarily passenger cars). The mutagenicity (TA 98; -S9) of the extractable particulate emissions from the spark-ignition vehicles was several times greater than that from the diesels, on a per  $\mu\text{g}$  of extract basis. But when mutagenicity was calculated on a revertants per km travelled basis, the mutagenicity of the particulate emissions from diesels exceeded that from gasoline-powered vehicles by approximately a factor of five.

They found that the diesel-produced aerosol at Allegheny was very similar to that encountered in dilution-tube studies with respect to percentage extractables, molecular-weight distribution, HPLC profiles, and particle size distribution. However, the extract mutagenicities in revertants/km for particulates collected from the dilution tube were 1.8 and 1.5 times higher for gasoline automobiles and heavy-duty diesels than the highway samples. Some of the difference between the dilution tube and highway results may be explained by the effect of road grade, wind and the tunnel site which decreases the particulate emissions from the heavy-duty diesels by 1.8 times (75).

The Allegheny Mountain Tunnel studies also served

Table 8. Comparison of emissions from laboratory and highway studies.

Parameter	Data by vehicle category					
	Automobiles-gasoline			Heavy-duty diesels		Light-duty diesels,
	Highway <sup>a,b</sup>	Dilution tube <sup>c</sup>		Highway <sup>a</sup>	Dilution tube <sup>c</sup>	Dilution tube <sup>c</sup>
		Noncatalyst	Catalyst			
Mass, mg/km						
Total particulates	18 $\pm$ 12	62 $\pm$ 19	10.6 $\pm$ 2.0	805 $\pm$ 54	1,036 $\pm$ 390	246 $\pm$ 6
Extractables	8.5 $\pm$ 3.5	9.5 $\pm$ 0.5	5.8 $\pm$ 0.2	189 $\pm$ 3	188 $\pm$ 44	124
Mutagenicity, rev/ $\mu\text{g}^d$						
TA 98 (-S9)	3.5 $\pm$ 0.5	6.4 $\pm$ 1.6	5.2 $\pm$ 2.3	0.75 $\pm$ 0.35	1.2	4.8 $\pm$ 2.3
TA 98 (+S9)	2.2 $\pm$ 0.2	13.0 $\pm$ 1.5	8.0 $\pm$ 3.0	0.6 $\pm$ 0.3	1.1	1.8 $\pm$ 0.5
Mutagenicity, rev/km						
TA 98 (-S9)	30	61	30	142	226	595

<sup>a</sup>Exhaust dilution ratio:  $10^3$ – $10^4$ ; average of May and August experiments. Average temperature  $16.5 \pm 4^\circ\text{C}$ .

<sup>b</sup>Ratio of noncatalyst to catalyst vehicles is approximately 1:1.

<sup>c</sup>Exhaust dilution ratio = 10–20. Average temperature =  $42 \pm 6^\circ\text{C}$ . From references (76–83) (weighted average of all data with variation given for 1 $\sigma$ ).

<sup>d</sup>mg of  $\text{CH}_2\text{Cl}_2$  extracted material.

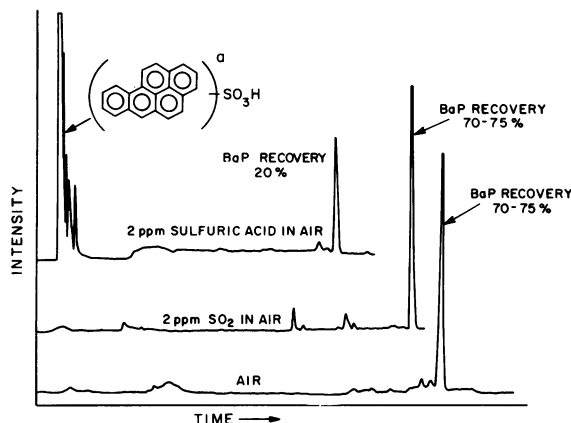


FIGURE 5. Products formed from the reaction of 500  $\mu\text{g}$  of BaP (deposited on T60A20 filter) with 2 ppm of  $\text{SO}_2$  or  $\text{H}_2\text{SO}_4$  for 30 min at a sampling rate of 20 L/min. Samples extracted with dichloromethane, analyzed on a 5  $\mu\text{m}$  Micropart  $\text{C}_{18}$  reverse phase column; linear gradient of 40%  $\text{CH}_3\text{CN}$  in water to 100%  $\text{CH}_3\text{CN}$  in 50 min.

as a sampling site for the determination of emission rates of 22 gas-phase hydrocarbons from motor vehicles in highway operation. Emission rates determined from that study proved generally consistent with tailpipe emission rates determined from published dynamometer studies (51).

## Summary

Nitro-PAH in diesel particulates are found to account for a major portion of the direct acting TA 98 Ames mutagenicity of diesel particulates. However, similar quantitative data for nitro-PAH are not yet available by using other *Salmonella typhimurium* strains and bioassay systems.

Very little is known about the formation of nitro-PAHs, sulfonated-PAHs, and other oxy-PAHs in the engine and tail pipe vehicle exhaust. More information concerning their chemistry and reaction kinetics under these conditions is needed.

It is difficult at this time to compare the mutagenic potential of vehicle exhaust with emissions from other sources since limited data exhaust with emissions from other sources since limited data are available. Qualitative and quantitative data on mutagens present in emission samples collected from a variety of other emission sources are needed.

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